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The Influence of Subsample Size in a Study of Freshwater Zooplankton

ABSTRACT

The influence of subsample size on counting precision and estimates of taxa richness is documented for a freshwater zooplankton biomonitoring program. Subsample variability was related to subsample size. The mean and median coefficient of variation for cladocera and copepoda were below 30% at counts of 50-100 individuals. Jaccard's similarity coefficient stabilized at counts of 100-150 individuals as did the total number of zooplankton taxa identified. These data suggest that counting more organisms in subsamples or more than one subsample per sample may do relatively little to better characterize samples.

Zooplankton studies typically employ a two-stage sampling scheme in which organisms are counted in aliquots drawn from field samples. Subsamples must be representative of the entire sample to estimate *in situ* densities accurately. Variability between subsamples decreases, i.e., counting precision increases, as the number of organisms in the aliquots gets larger (Langeland and Rognerud 1974, Alden et al. 1982, Lund 1999, Graves 2004). Estimates of taxa richness also increase with subsample size (Fisher et al. 1943, Graves 2004). However, the gain in precision or taxa richness decreases as subsample size (and counting effort) increases (e.g., Graves 2004) and at some point may not justify the extra cost of analysis. Guidance on the appropriate subsample size for zooplankton studies is often based on theoretical or limited experimental data (e.g., Downing and Rigler 1984, APHA 1998). Few comparative data have been published that document the influence of sample size in practice. This paper presents such an analysis from a QA/QC program associated with a multi-year biomonitoring program.

Zooplankton were collected monthly for 4 years from a cooling reservoir and downstream wetland located on the Department of Energy's Savannah River Site in South Carolina using an 80 µm mesh plankton net and bucket which retained all cladocera, copepoda and larger rotifer species. A Stempel pipette was used to obtain subsamples. Usually, only single subsamples were enumerated. A second subsample from 226 randomly selected samples (approximately 10% of all samples collected) was counted by the same technician to assess subsampling precision. Most zooplankton were identified to genus or species; immature copepods were categorized as nauplius larvae, cyclopoid copepodids or calanoid copepodids. Multiple technicians counted subsamples. The influence of subsample size was quantified using the coefficient of variation (CV) between pairs of subsamples for counts of cladocera and copepoda, the total number of taxa identified in both subsamples and Jaccard's similarity coefficient (Krebs 1999):

$$S_j = \frac{a}{a+b+c}$$

where a = number of taxa in both subsamples, b = number of taxa in subsample A but not in B and c = number of taxa in subsample B but not in A.

Subsample variability was related to subsample size. The CV for cladocera and copeopda decreased, while Jaccard's similarity coefficient increased, with increasing organism counts (Fig. 1). Subsampling error up to 30% is considered reasonable for zooplankton studies (Langeland and Rognerud 1974, Alden et al. 1982, Lund 1999). The mean and median CV for both cladocera and copepoda fell below this threshold at counts of 50-100 individuals. Jaccard's similarity coefficient stabilized at counts of 100-150 individuals as did the total number of zooplankton taxa. Recommendations for subsample size in zooplankton studies vary from > 25 (de Nie and Vijverberg 1985) to 200-500 organisms (APHA 1998). Data from this and other studies (e.g., Alden et al. 1982 and Downing and Rigler 1984) suggest that only minimal gain in precision or sample information is achieved by counting more than 100-150 individuals in subsamples. Guided by preliminary QA/QC results, samples during this study were concentrated so each subsample contained at least 75-100 organisms. Provided there is adequate precision, counting only single subsamples allows for the reallocation of resources (time and money) to collecting and processing more field samples.

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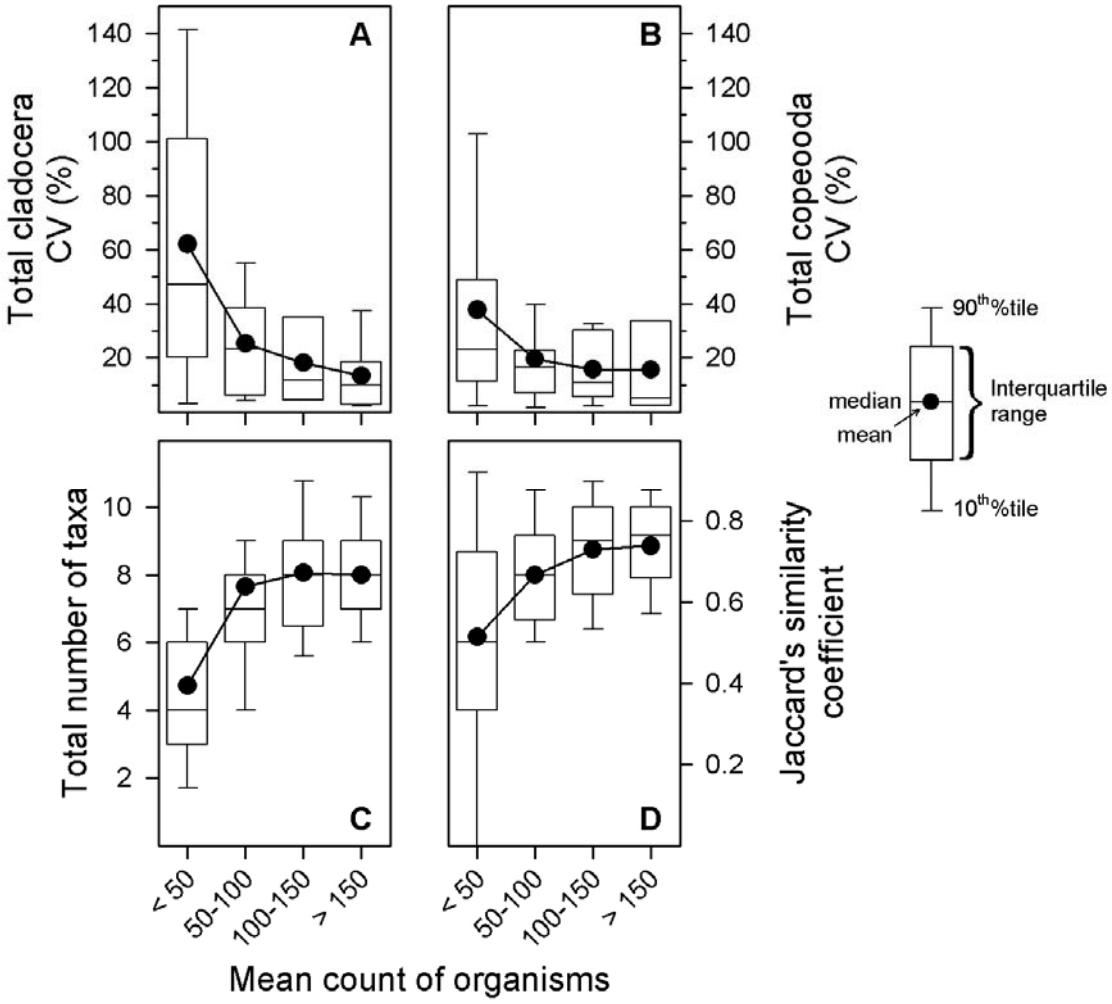

Figure 1. Relationship between mean counts of individuals in pairs of subsamples and (A) coefficient of variation (CV) for counts of cladocerans, (B) CV for copepods, (C) total number of zooplankton taxa identified in both subsamples and (D) Jaccard's similarity coefficient.